

## CLAIMS

***What is claimed is:***

1. A method of investigating a treatment applied to a plurality of cells, the treatment having at least an on-target effect on the plurality of cells, the method comprising:  
identifying at least an on-target cellular feature or group of on-target cellular features of the plurality of cells, the on-target cellular feature or features being affected by the treatment and being related to the on-target effect;  
identifying at least an off-target cellular feature or group of off-target cellular features different to the on-target cellular feature or features, which are also affected by the treatment and which are related to a side effect of the treatment; and  
determining a measure of the side effect based on the off-target cellular feature or features.
2. The method as claimed in claim 1, further comprising characterising the treatment based on the measure of the side effect.
3. The method as claimed in claim 1, further comprising determining a measure of the on-target effect based on the on-target cellular feature or features.
4. The method as claimed in claim 3, further comprising characterising the treatment based on the measure of the on-target effect.
5. The method as claimed in claim 4, further comprising characterising the treatment based on the measure of the side effect and the measure of the on-target effect.
6. The method as claimed in claim 1, wherein the off-target cellular feature or features are not related to the on-target effect.
7. The method as claimed in claim 1, wherein the measure is a distance in a multivariate space corresponding to the off-target cellular features.

8. A method of characterising a treatment that has been applied to a population of cells and that has an on-target effect on the population of cells, comprising:
  - identifying from a plurality of cellular features of the population of cells, a first group of cellular features which have been affected by the treatment and which are related to the on-target effect of the treatment;
  - identifying from the plurality of cellular features a second group of cellular features which have been affected by the treatment and which are not related to the on-target effect of the treatment;
  - creating a first signature characteristic of the on-target effect from the first group of cellular features;
  - creating a second signature not characteristic of the on-target effect from the second group of cellular features; and
  - evaluating a first measure derived from the first signature and a second measure derived from the second signature to characterise the treatment.
9. The method as claimed in claim 8, and further comprising:
  - determining the separation in multivariate space between the second signature and an origin.
10. The method as claimed in claim 9, further comprising:
  - determining the separation in multivariate space between the first signature and an origin.
11. The method as claimed in claim 9, wherein the origin is provided by a control signature created from a control group of cellular features of a control group of cells, and wherein the control group of cellular features are the same cellular features as the second group of cellular features.
12. The method as claimed in claim 10, wherein the origin is provided by a control quantitative signature created from a control group of cellular features of a control group of cells, and wherein the control group of cellular features are the same cellular features as the first group of cellular features.
13. A computer program product comprising a machine readable medium on which is provided program instructions for characterising a treatment that has been applied

to a population of cells and that has an on-target effect on the population of cells, the instructions comprising:

code for identifying from a plurality of cellular features of the population of cells, a first group of features which have been affected by the treatment and which are related to the on-target effect of the treatment;

code for identifying from the plurality of cellular features a second group of features which have been affected by the stimulus and which are not related to the on-target effect of the treatment;

code for creating a metric characteristic of the on-target effect from the first group of features;

code for creating a second metric not characteristic of the on-target effect from the second group of features; and

code for evaluating the first and second metrics to characterise the treatment.

14. A computing device comprising a memory device configured to store at least temporarily program instructions for characterising a stimulus that has been applied to a population of cells and that has an on-target effect on the population of cells, the instructions comprising:

code for identifying from a plurality of cellular features of the population of cells, a first group of features which have been affected by the treatment and which are related to the on-target effect of the treatment;

code for identifying from the plurality of cellular features a second group of features which have been affected by the treatment and which are not related to the on-target effect of the treatment;

code for creating a first metric characteristic of the on-target effect from the first group of features;

code for creating a second metric not characteristic of the on-target effect from the second group of features; and

code for evaluating the first and second metrics to characterise the treatment.

15. A method of characterising a treatment applied to a population of cells, comprising:  
deriving a plurality of cellular features from at least a first captured image of the population of cells that have been exposed to the treatment;

creating an on-target effect signature, which is characteristic of an on-target effect of the treatment on the population of cells, from at least a first one of the plurality of cellular features, the at least one of the plurality of features relating to cellular properties involved in the on-target effect;

creating a side effect signature, which is characteristic of a side effect to the on-target effect, from at least a second one of the plurality of cellular features, the second one of the plurality of cellular features relating to cellular properties not being involved in the on-target effect; and

evaluating an on-target effect metric derived from the on-target effect signature and/or a side effect metric derived from the side effect signature to characterise the treatment.

16. The method as claimed in claim 15, wherein the on-target effect signature is created from a group of cellular features.

17. The method as claimed in claim 16, wherein the side effect signature is created from a further group of cellular features, in which none of the members of the group of cellular features used to create the on-target effect signature and the members of the further group of cellular features used to create the side effect signature are common.

18. The method as claimed in claim 15, wherein the second one of the plurality of cellular features is affected by the treatment.

19. The method as claimed in claim 18, further comprising:  
exposing different populations of cells to different doses of the treatment; and  
deriving the on-target effect metric and the side effect metric for different doses of the treatment.

20. The method as claimed in claim 15, wherein deriving the on-target effect metric or the side effect metric includes determining the difference between the on-target effect signature or side effect signature and a control signature from the same cellular features for a control group of cells.

21. The method as claimed in claim 15, and further comprising:  
capturing at least a first image of a control group of cells; and  
deriving a plurality of cellular features from the image of the control group of cells;  
creating a control on-target signature for the same cellular features for the control group; and  
creating a control side effect signature for the same cellular features for the control group.
22. The method of claim 21, further comprising determining a side effect distance in a multivariate space between the side effect signature and the control side effect signature.
23. The method of claim 22, further comprising determining a target effect distance in a multivariate space between the on-target effect signature and the control on-target effect signature.
24. The method of claim 23, wherein characterising the stimulus is based on the side effect distance.
25. The method of claim 24, wherein characterising the stimulus is based on the on-target effect distance.
26. The method as claimed in claim 25, further comprising generating a graphical representation of the side effect distance and on-target effect distance.